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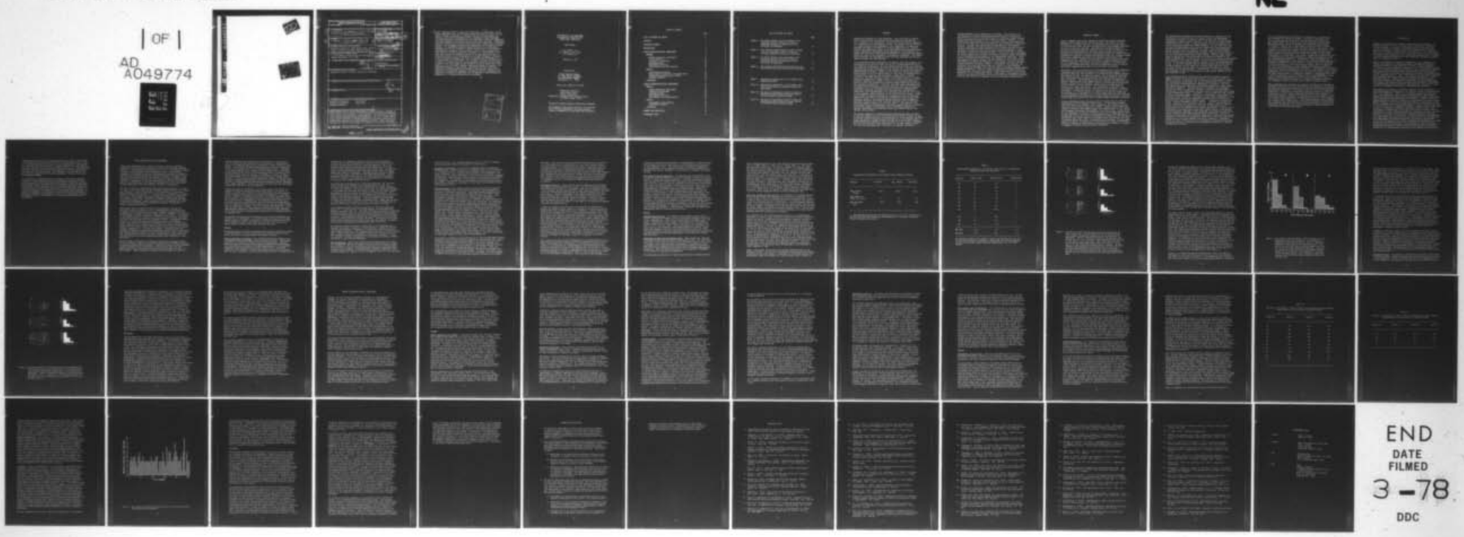

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This document reports initial development of ~~two~~ (2) experimental programs to permit quantitative assessment of the feasibility of Acupuncture Analgesia under clinical and especially field conditions. One experimental program involved Acute Neurophysiological Experiments in the cat to permit quantitative characterizations of optimal Acupuncture waveforms (electrical) or manipulations (manual) and optimal Acupuncture points. These experiments involved the study of the influence of Acupuncture stimulation on single
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neuron responses driven by tooth pulp stimulation in ~~two~~ 2 thalamic nuclear groups associated with the affective-motivational aspects of pain. At the time of contract termination, techniques and facility development were complete, pulp-elicited thalamic unitary responses had been catalogued to establish general characteristics, and preliminary definitive experiments involving simulated Acupuncture stimulation had been completed. The results show that stimulation of the Hoku Acupuncture point significantly attenuates thalamic unit activity elicited by pulp stimulation in both nuclear groups studied. The ~~second~~ 2nd experimental ~~program~~ involved chronic Behavioral experiments in the cat to permit a direct verification of the feasibility of Acupuncture analgesia at the perceptual level to avoid interpretive complications of placebo reactions, hypnotism, etc. Development of this model would permit the immediate testing of the perceptual efficacy of conclusions from the acute experiments, and by the placement of temporary blocks or permanent lesions, the study of pathways and mechanisms to aid in optimization of effectiveness. At the time of contract termination, all procedures of animal selection, chronic surgical implantation, and animal training to the final stage of the Threshold Titration program were operational, as was an Automatic Shaping electronics system for control of all experimental stages in the program. Preliminary Threshold Titration protocols involving Acupuncture stimulation qualitatively indicated the induction of recognizable analgesia. Premature contract termination prevented the collection of sufficient data to adequately assess Acupuncture analgesia or to optimize administration techniques.

ACUPUNCTURE IN THE MANAGEMENT
OF INJURY AND OPERATIVE PAIN
UNDER FIELD CONDITIONS

FINAL REPORT

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ABSTRACT

Acupuncture analgesia is a real phenomenon. This conclusion is inescapable, in spite of wide vacillations in the popularity of Acupuncture (to be expected with revolutionary procedures) and complications regarding the interpretation of experimental results. The present work was directed to a rigorous quantitative assessment of the feasibility of Acupuncture analgesia and means to optimize its effective administration. The work involved two experimental series, Acute Neurophysiological Experiments and Chronic (Behavioral) Neurophysiological Experiments. Unfortunately, the program was terminated following the development of appropriate experimental models but prior to the realization of definitive conclusions. The present report describes the experimental models that were developed and the preliminary results that were obtained.

Acute Neurophysiological Experiments were based on three key elements, pulp stimulation as the source of pain, the use of the Hoku Acupuncture point for Acupuncture stimulation, and the attempted identification of a specific recording site in the brain, the activity of which was to be used as an index permitting quantitative assessment of the efficacy of particular Acupuncture administration strategies. Tooth pulp was chosen for noxious test stimulation because it is a recognized source of pure pain, is representative of orofacial pain in general, and represents a tissue which is readily accessible. The Hoku Acupuncture point was selected because of its popularity concerning pain of dental origin. As an index of activity in pain pathways, responses were sought in the area of the thalamus which were driven by tooth pulp stimulation and whose responsiveness was affected by the Acupuncture stimulation. Initially, the Acute animal preparation was developed and field-potential recording techniques were used to delineate the general area of the thalamus exhibiting pulp-driven responsiveness. Single-unit recording techniques were then developed, and employed to characterize the responsiveness of the individual pulp-driven units found in three thalamic areas identified in the field-potential studies. Finally, the Dot-Raster recording technique was introduced and used to rigorously characterize the effects of Acupuncture stimulation on pulp-driven units primarily located in the posterior nuclear group but also in regions of the medial thalamus, both areas primarily associated with the emotional-affective aspects of pain (i.e., the real 'hurt'). The results of the preliminary definitive experiments conclusively demonstrated the feasibility of the model system; the particular form of Acupuncture stimulation employed consistently and reversibly reduced the responsiveness of the thalamic units studied.

The Chronic (Behavioral) Neurophysiological experimental model was based on four key elements, the use of tooth pulp as the source of pain, the use of the Hoku Acupuncture point, the implantation of a head pedestal providing electrical access to animal implants, and the use of a perceptual experimental paradigm (as opposed to using an index of reflex or motor responses). The tooth pulp and Hoku points were chosen for the same reasons as given for the Acute Experiments and, furthermore, to maximize information

exchange between the two experimental procedures. Construction of the head pedestal was adapted from procedures described in the literature, and was found to be the only adequate means to establish reliable periodic electrical connections between the animal and external equipment. It was deemed absolutely necessary to use a perceptual index of pain sensibility, as it is well known that reflex responses are poor predictors of perceptual efficacy. The particular perceptual paradigm employed, the Threshold Titration paradigm, was adopted because this procedure permitted the quantitative monitoring of pulpal thresholds over prolonged periods of time. Initially, we developed the surgical model, and then proceeded to develop the animal facility and training procedures. During this time, requirements for mutual compatibility dictated considerable interplay between development of the preparation and development of the experimental facilities. Subsequently, a Threshold Titration module was designed and fabricated, surgery was conducted on several animals, and the feasibility of all training and experimental procedures was documented. From program initiation, Automatic Shaping procedures (automatic conduct of experiments) were employed, and eventually the system was placed under the control of a microprocessor (previous control equipment which had been used for Automatic Shaping suddenly became unavailable). Finally, a number of preliminary Threshold Titration procedures were conducted, the results of which quantitatively verified the feasibility of our experimental design. Nevertheless, the premature termination of the contract precluded collection of sufficient definitive Threshold Titration data relating to analgesia to permit conclusive interpretation.

CONCEPTUAL SUMMARY

Local or regional pain control agents and techniques presently available possess certain drawbacks and contraindications. Nerve block or local infiltration is usually effective, but disadvantages include long induction and recovery times, and such contingencies as tissue damage, infection, and toxic or allergic reactions. Inhalation or intravenous agents for general anesthesia provide more extensive control of pain and anxiety, but potentiate the disadvantages of the localized procedures and introduce prolonged perturbations of various physiological parameters, General Electroanesthesia has undergone extensive international study, and, although plagued with certain drawbacks, may finally be exhibiting potentialities for manageable utility. Regional Electroanalgesia, a new procedure under investigation which shows much promise, has not yet reached operational status. The electroanalgesic techniques, both local and general, are attractive, because most problems which have continuously hampered the use of injected and inhaled pharmacological agents can be directly circumvented.

Acupuncture, like the various forms of Electroanesthesia or Electroanalgesia, circumvents most problems encountered with pharmacological agents. As previously mentioned, the instruments of Acupuncture are extremely portable, and allergic or toxic reactions are unknown. The technique has potential use, *with limitations*, by nonprofessional aide-level personnel under emergency conditions. Using mechanical induction (twirling the Acupuncture needle), infection and tissue damage remain as realistic contingencies, but the much more attractive non-invasive electrical stimulation of Acupuncture points is likely to prove feasible, thereby totally circumventing all necessity for tissue penetration. Also, Acupuncture blocks pain, not consciousness, permitting patient cooperation during transport or surgical procedures. In addition, recovery is reported to be uneventful, there is no equivalent to a drug hangover, all senses except pain are normal, and vital physiological parameters are not only uncompromised but supposedly are actively maintained both during and following surgical procedures. Possibly, certain high-risk patients can only be treated using Acupuncture procedures.

Certain disadvantages have also been noted for Acupuncture, but they are overwhelmed by the advantages mentioned above. In many cases extended induction periods are required. Also, Acupuncture is ineffective in some people and only partially effective in others. Finally, some studies have revealed a possible relationship between the "suggestibility" of the subject and subsequent Acupuncture effectiveness. The latter point, if true, would not be surprising, because the effectiveness of any analgesia agent includes placebo components, at least at certain concentrations. Nevertheless, it is important to note that there is absolutely no evidence for any neurological differences which might subserve the abovementioned contingencies, and such differences may be the result of inadequacies of technique. In this regard, present Acupuncture procedures are not only diverse but quite inconsistent. Therefore, Acupuncture shows marked potentialities as, minimally, an adjunct to analgesic procedures enjoying present popularity.

The physiological mechanisms underlying Acupuncture are unknown, and even descriptions of the numbers and locations of "points" are undergoing contemporary evolution. Therefore, traditional practical and conceptual information is highly suspect. Because of this fact and to insure optimal potentialities for success, a balanced approach along three distinct but highly interrelated avenues was deemed mandatory. To the latter end, both an Acute Neurophysiological and a Chronic (Behavioral) Neurophysiological experimental series were concurrently initiated. These two experimental series in animals were felt essential to permit rapid sorting of experimental variables and to generate data necessary to obtain official sanction for the third experimental series, Human studies. Eventual Human studies would be required to verify conclusions and techniques developed in the animal investigations. Tooth pulp was to be used as the source of experimental pain, because it was easily accessible, it was classically considered to be a source of pure pain, and was readily adaptable to all three experimental series. It was also intended that all primary Acupuncture points enjoying maximal popularity for the control of orofacial pain (there are three; see below) would be tested singly and in various combinations to insure effective and thorough evaluation. Unfortunately, at the time the project was terminated, it had only been possible to look at data related to the Hoku Acupuncture point.

Acute Neurophysiological Experiments were required to identify effective animal Acupuncture points, and, subsequently, to optimize the effect in terms of "points" and waveforms and to establish the maintenance of reasonable physiological homeostasis (for safety considerations in conjunction with the Chronic Neurophysiological Experiments in animals). The Acute experiments would also permit the quantitative comparison of manual versus electrical Acupuncture. Although the above information could be gained from Chronic models, it was felt essential to include Acute experiments, because many of the experimental variables could be sorted much faster using an Acute experimental model. For example, the long induction times involved in Chronic experiments make it difficult to investigate a large number of variables in each experiment. Furthermore, Acute animal experiments provide more flexibility in terms of permissible waveforms and in the ability to accurately and quantitatively identify effective activation of Acupuncture points. Chronic experiments lack an effective criterion for Acupuncture "point" activation, and, therefore, would not provide a means to evaluate whether unsuccessful findings were the result of ineffective "point" activation or if the Acupuncture was actually ineffective. The Acute Neurophysiological data would also be much more closely related to the general neurophysiological knowledge of pain mechanisms, and therefore would permit better correlation with previous physiological information. The basic approach employed single-unit recording techniques in specific brain centers suspected or identified as providing an index of the effects of Acupuncture stimulation on tooth pulp pathways involved in perception. Eventually, certain of these protocols were to be accompanied by continuous monitoring of various indices of cardiovascular and metabolic parameters, to provide documentation of safety.

Chronic Neurophysiological Experiments in animals were to provide the main framework for the entire program. The Chronic experiments would provide a key link between the Acute Neurophysiological studies and the ultimate Human work, because the Chronic Neurophysiological Experiments would permit tracing of pathways and mechanisms (using chronic recordings or lesions) in the same animal in which behavioral effects of Acupuncture stimulation on pain perception had been quantitated. Also, behavioral situations would permit the effectiveness of Acupuncture to be examined free from placebo effects, distraction, hypnotism, and various other potential contributing factors which are plaguing present Human investigations of Acupuncture. The Chronic studies were required initially in conjunction with the Acute experiments to identify Acupuncture points and to demonstrate feasibility. Subsequently, these experiments were considered indispensible to test the use of the specific Acupuncture points in various combinations and the effects of various stimulation waveforms on actual perception, to test psychological effects, and, in conjunction with the Acute experiments, to study optimal administration parameters and verify safety and physiological homeostasis. Fundamentally, the Chronic preparation involved simultaneously the use of chronic implants for electrical stimulation of the test tooth pulp and of specific Acupuncture points, with the animal being tested in a behavioral paradigm for <u>perceptual</u> sensitivity to pain (as opposed to reflex flinching or other motor responses). Eventually, certain experiments were to have been accompanied by chronic recording from various brain centers, by repeated experiments following the placement of specific lesions, and by tests to document safety.

Human experiments were scheduled for eventual inclusion in the program to provide the ultimate test of all physiological, psychological, perceptual, and application characterizations derived from the Acute and Chronic Neurophysiological Experiments. We planned to test the ability of Acupuncture stimulation to attenuate the perceptual experience elicited by electrical stimulation of intact teeth, using Signal Detection Theory formats of experimental design to separate true changes in sensory information inflow to the brain from alterations in response criteria of the subject (mental set, anxiety level, hunger, etc.). Such Human experiments were programed for initiation after the majority of the Acute and Chronic experiments had been completed; the latter goal had not been accomplished at the time the program was terminated, so no further discussion of human experimentation is included in the present discussion.

INTRODUCTION

Analgesia can definitely be produced using Acupuncture techniques. Acupuncture was used for over a thousand years for therapeutic treatment in Asia, although, even there, its analgesic potentialities were just recently discovered. Although contributing phenomena such as placebo reactions, distraction, conditioning, or hypnotism (1-6) may occasionally play a role, careful clinical (1,7-11) and laboratory (1,2,12,13) studies in humans and the demonstration that Acupuncture Analgesia works in animals (1,14-16) establishes unequivocally that Acupuncture treatment strategies exert true analgesic effects.

Albeit it is known that Acupuncture Analgesia is a true phenomenon, the concepts which embody our traditional medical knowledge and theories have been deficient in their abilities to offer a logical mechanistic hypothesis to explain Acupuncture Analgesia (7,17). Nevertheless, Acupuncture is receiving considerable international attention spurred by purported advantages that include portability, infinite shelf-life, and complete freedom from toxic or allergic contingencies associated with present pain-control techniques (15,17). Also, cardiovascular and respiratory parameters are purportedly unaffected, which is particularly desirable in certain high-risk patients. Finally, electrical Acupuncture stimulation, an attractive alternative to mechanical stimulation, is easily administered and requires less attention than previously used anesthetics. This method of Acupuncture may afford noninvasive application (4) and potential opportunities for emergency application by nonprofessional personnel.

Our program was directed to a rigorous evaluation of the feasibility of employing Acupuncture Analgesia for the control of injury or operative pain under field conditions. Initially, the program involved the concurrent use of two experimental models, Acute Neurophysiological Experiments in animals and Chronic Neurophysiological Experiments in animals, respectively. Acute Neurophysiological Experiments were required to identify effective animal Acupuncture "points", to permit the rapid sorting of various experimental variables, and to define permissible limits of stimulation for the Chronic experiments and eventual Human studies. Many of these factors were the direct result of the increased flexibility afforded by Acute experiments, in terms of permissible stimulation parameters, enhanced control over experimental variables, and the fact that quantitative criteria could be established to verify effective activation of Acupuncture points. The basic approach was to utilize well-established neurophysiological recording techniques, described in detail below. Chronic (Behavioral) Neurophysiological Experiments were to provide the key link between Acute Neurophysiological Experiments and Human investigations, because they would permit collection of definitive data related to pathways and mechanisms in the same animal in which behavioral effects of Acupuncture stimulation on pain perception could be quantitated. Chronic experiments were also valuable because they avoided the complications of placebo reactions, hypnotism, distraction, or conditioning, and permitted, in combination with the Acute

experiments, the definition of permissible limits of safety. The validity and applicability of the Chronic model was to be established first. This had just been completed at the time of project termination. Subsequently, the model was to be used to identify optimal waveforms for three critical Acupuncture "points" associated with orofacial pain control. The same model was then to be used to study the three Acupuncture points in various combinations and to document safety. More details of the rationale of the dual approach using Acute and Chronic experiments in animals is given in the Conceptual Summary.

The combination of complementary and confirmatory information which was scheduled to be obtained from these two experimental series would have provided a powerful information base upon which the feasibility and potential ultimate utility of Acupuncture as an analgesic technique could have been defined. This combination would also have provided data essential to obtain sanction for quantitative evaluation of the therapeutic utility of Acupuncture Analgesia in humans and to solve bioengineering problems. The untimely termination of the program at the point where both experimental models were completely operational but short of the significant collection of definitive data on the feasibility of Acupuncture analgesia was very unfortunate.

ACUTE NEUROPHYSIOLOGICAL EXPERIMENTS

Present data establishes that there is a definite analgesia component to Acupuncture, over and above contributing factors such as placebo reations, distraction, conditioning, and hypnotism (1,2,7-17). In our overall program directed to the assessment of the therapeutic utility of Acupuncture Analgesia, Acute Neurophysiological Experiments were required to permit the flexibility of physical access and recording techniques not readily available in Chronic experiments and unaccessible for studies in humans. Acute experiments also were to permit the rapid survey of multiple experimental variables. Fundamental to the Acute experiments was the fact that Acupuncture has also been shown to be effective in animals (1,14-16), data which further substantiates the true analgesic efficacy of Acupuncture.

The basis of the Acute experimental model was the identification of a locus (or loci) in the brain which had the following characteristics: a), activity was indicative of noxious stimulation of the tooth pulp; b), there was a suspected direct participation in the transmission of information to perceptual (not reflex) centers; and, c), activity was attenuated by Acupuncture stimulation. This general experimental design was schematically depicted in a previous report (18). Activity from thalamic units meeting all three criteria were designated "index" responses, and such responses, once identified, were to be employed to quantitatively evaluate the effects of specific Acupuncture protocols.

Experimental pain was induced by tooth pulp stimulation, because this structure is readily accessible (19), is representative of orofacial pain in general (20), and because the perceptual response in humans (19,21,22) and apparently in animals (23,24) is always painful, regardless of the nature of the stimulus (thermal, mechanical, chemical, or electrical). Furthermore, it was chosen because of correlation with previous data from the literature (20,25-35) and from work within our laboratory (36-42). Rectangular stimulus pulses of 0.1 ms duration and suprathreshold intensity were employed. Stimuli were applied in the bipolar configuration to insure limitations of current within the pulp chamber proper (37,43), and constant current stimulus output units were used to insure uniform stimuli in the event of alterations of pulpul impedence (44). A short train (burst) of stimulus pulses was employed, as opposed to single shock stimulation, because there is significant cant evidence from the literature (45-47) and especially from our laboratory (48) that effective activation of units at high levels in the central nervous system elicited by pulp stimulation exhibit lower thresholds to train stimulation.

The best "index" indicative of nociceptive activity and Acupuncture interactions is unknown. However, a reasonable approximation was made based upon the available physiological knowledge. There are at least two major ascending systems participating in the affective or motivational aspects of pain, heavily colored by a third pathway which modulates the other two (46). All three pathways exhibit anatomical proximity in the thalamus, but involve

discreet anatomical and physiological nuclear groups in that structure. Also, all three thalamic nuclear groups are located at a high level of integration, permitting many prior interactions to have occurred at lower levels. A few studies had implicated Acupuncture effects on nociceptive units in the thalamus prior to our initial proposal (49-51). Therefore, our proposed approach was to examine in detail single units in all three nuclear groups at the level of the thalamus involved in nociception and choose one nuclear group as the best, if possible, to use as an index of the effectiveness of various strategies of Acupuncture Analgesia administration. At the time of contract termination, however, it had only been possible to examine representative neurones of the posterior nuclear group (PO) and the Centrum Median-Parafascicular-Intralaminar complex (designated MT for medial thalamus). Units of the third nuclear group, the ventrobasal complex (VB), had not as yet been examined.

Regarding Acupuncture stimulation, our original plan was to look at all acupuncture points associated with dental pain relief. Initially, we looked at the Hoku point because of its extreme popularity; the premature project termination precluded the systematic investigation of any other points. Actually, we stimulated a point on the relevant peripheral nerve just proximal to the Hoku point; this procedure has been shown to be similar in its effects to stimulation of the Acupuncture point itself (17). An optimal electrical waveform based upon the literature and information from our laboratory was selected to search for our central nervous system index of Acupuncture efficacy. Had such an index been identified, it would then have been used to optimize Acupuncture points, waveforms, and other administration parameters. Unfortunately, the definitive data available at the time of project termination was insufficient to conclusively define the desired index.

Standardized neurophysiological stimulation and recording techniques were employed for all experiments. The anesthetized cat was used as the experimental animal for economy, availability, correlation with previous work in our laboratory (36-42), and based upon the wide use of the cat as a general model of somatic (26,52) and orofacial (25) pain.

Methods

The methodological particulars presented below have been described in detail in previous reports (18,53). The material that follows includes considerable repetition for completeness of the present document.

Animal Preparation and Surgery. The acclimated animal was brought to the experimental surgery and premedicated with atropine sulfate. Endotracheal entubation was accomplished using a short-acting barbituate. Surgical anesthesia was induced using Ethrane-Oxygen and maintained during surgery and the experimental procedures using Ethrane-N_2O-Oxygen. This anesthetic combination was employed because it mimics the effects of Chloralose, classically, the anesthetic of choice in studies of activity in pain pathways (54) and in studies of central nervous system effects of Acupuncture (55).

Subsequently, an esophogeal temperature probe and stethescope were inserted, an optically-coupled ECG (to preserve ground isolation) was attached (Terrasyn Model N-IIIB ECG Isolation Amplifier), and a non-invasive blood-pressure monitoring system was applied to one of the anterior limbs (Hoffman-La Roche Arteriosonde, Model 1010). In addition, short-term acid-base balance (end-tidal CO_2) was monitored utilizing a Bechman LB-2 Medical Gas Analyzer. Respiration was assisted on demand, or controlled, using a Bird Mark IV-VIII Anesthesia Assistor. Intra-esophogeal temperature was recorded (YSI Model 43TA) and maintained at 38 $\pm$ 0.5 C by means of a heating pad with thermostatically controlled circulating water (Gaymar Temp-Pump System).

The animal was first mounted in a stereotaxic apparatus, and two stimulation electrodes were placed in the experimental tooth (37). One electrode was placed in each of two cavities drilled through the enamel to a near pulp exposure. The base of each cavity was filled with Eccobond, the electrode wire (stranded) was packed in place, and the whole complex was sealed in position using a non-conductive filling material. One stimulus electrode was located on the lingual aspect and one on the buccal aspect of the maxillary canine test tooth, each electrode electrically isolated from the gingival margin. Electrode materials were platinum to reduce the effects of electrode polarization (44). Based upon our data (37) and data from the literature (43,56), the stimulus configuration as described effectively limits current flow to the test tooth pulp.

All experiments involved bilateral stimulation of the Hoku Acupuncture points (57). The Hoku point is the most popular site associated with maxillary orofacial pain (57). Each Hoku electrode was placed by surgically exposing the medial branch of the superficial radial nerve just proximal to the actual Hoku point and wrapping the bared stimulus lead wire around the nerve. The ipsilateral electrode was the cathode (58), and the stimulation circuit was completed through the animal. Stimulation of the peripheral nerve supplying the Acupuncture point has been shown to be equally as effective as stimulating the Acupuncture point itself (17).

The surgical procedure required to gain access to the recording site involved the initial reflection of cranial muscles to expose the cerebrum overlying the thalamus. Subsequently, a defect was created in the hard tissue using a dental bur. The resultant defect was then filled with saline agar to afford protection from exposure and to limit artifacts from cerebral movements induced by circulatory or respiratory pressures. It was not necessary to create a pneumothorax or to ventilate particular cerebral ventricles to limit cerebral movements.

Pulp Stimulation. Noxious test stimulation was administered in the bipolar configuration between the lingual and buccal pulp electrodes. Stimuli were delivered to the two platinum test electrodes from a battery-powered constant-current generator (lab-constructed), triggered by and photically-isolated from one channel of a Grass S-88 Physiological Stimulator. Each stimulus episode was composed ot three rectangular pulses of 0.1 ms duration

applied at 300 Hz. Test stimulus intensity was set at 150% of threshold tor the particular pulp-driven thalamic unit under study.

Acupuncture Stimulation. The electrical waveform used for Acupuncture stimulation was a continuous train of rectangular pulses of 0.1 ms duration at a frequency of 50 Hz. Stimuli were delivered to the stimulation electrodes (platinum) from a battery-powered constant-current generator (lab-constructed), in a manner analogous to the tooth pulp stimulation, triggered and photically isolated from a second channel of the Grass S-88 Physiological Stimulator. The intensity for stimulation at the Acupuncture points was slightly (ten percent) below that necessary to induce visible muscular fasiculation, typically on the order of 1 ma, which correlates with values used clinically (7,57).

Recording System. All recording protocols were initiated by a systematic search of the particular nuclear group in question, using standardized stereotaxic techniques (see below). Preliminary mapping experiments were conducted using field-potential recording techniques and Frederick Haer Epoxy-Coated stainless steel bipolar concentric macroelectrodes. Using the anatomical domains defined by the field-potential experiments, preliminary experiments were conducted to establish the characteristics of single units in identified thalamic areas, and then definitive Acupuncture protocols were conducted using the single-unit recordings. The electrodes employed were Frederick Haer Epoxy-Coated Tungsten Wire Microelectrodes, having a tip diameter less than 1 μ and an exposed tip not exceeding 5 μ. The recording system involved, sequentially, a Frederick-Haer d.c. coupled preamplifier postioned near the animal, a Mentor F-60 active 60 Hz notch filter, and a 3A9 differential preamplifier (in a Tektronix 129 4-Bin Power Supply). Three channels originated from the 3A9, an audio signal derived from a Frederick-Haer Audio Analyzer, unitary response waveforms with superimposed discriminator window levels derived from a Frederick-Haer Amplitude Analyzer (window discriminator) displayed on channel one of a Tektronix 3A74 mounted in a 564-B oscilloscope, and a dot-raster display derived from the same Frederick-Haer Amplitude Analyzer to a lab-constructed raster generator and displayed on channel two of the Tektronix 3A74 in the 564 B oscilloscope. Film records of the dot-raster data were obtained using a Nihon-Kohden PC-2A scope-mounted camera. The dot-raster display format involved time on the x axis, trial number on the y axis (by virtue of sweep position determined by the raster stepper), and a dot representing an individual response on the z axis (z-axis modulation).

As mentioned, the method of data acquisition for the Acute Neurophysiological Experiments relied on the dot-raster technique of data display (59) recently introduced into neurophysiological research. The central concept of the dot-raster technique lies in the fact that in recording responses from single units, knowledge of the occurrence of the unitary response is sufficient, and information regarding waveform is not relevant. Using the dot-raster technique, an individual stimulus episode was represented as a single horizontal sweep on the oscilloscope. No signal was present unless a response occurred, but, each time the unit responded, a dot was displayed at that point in time on the oscilloscope sweep to represent the neuronal

discharge. This procedure was accomplished by modulating the intensity of the oscilloscope beam (z-axis modulation). The first stimulus episode (individual sweep) of an extended sweep-stimulus presentation series was presented near the top of the oscilloscope screen. Subsequent stimulus episodes were presented as additional horizontal sweeps positioned at successively lower levels on the oscilloscope screen (by the raster-stepper). This technique afforded the ability to present and store for photography many (typically, 64) stimulus episodes on a single oscilloscope screen. This sequence of data collection and recording allowed the compilation of voluminous data into a form that was readily available and easily interpreted.

Experimental Protocol. Three distinct nuclear complexes of the thalamus are presently associated with pain (45,46). All Acute Neurophysiological Experiments were initiated by a systematic search of the requisite contralateral thalamic nuclear group (PO, MT, or VB), using stereotaxic techniques based upon our experience and information from the literature (26, 54). As mentioned, following development of the animal preparation and experimental equipment array, preliminary field-potential experiments were conducted to delineate thalamic regions of interest. Details of the preliminary mapping procedures and results are given in a previous report (18), and were essentially similar to that used for the single-unit mapping techniques as described below.

A search for single pulp-driven units was conducted by systematically mapping the thalamic nuclear complex of interest in vertical tracts along medial-lateral and proximal-distal grid coordinates. A given penetration involved the extremely slow (e.g. 1-2 μ/s) manual advancement of the electrode tip in the vertical tract during stimulation of the test pulp, using the standard test stimulus parameters (see above). Once a pulp-driven unit was located, an attempt was made (cautiously) to optimize response amplitude, and then the physiological properties of the unit were characterized. The parameters which were quantitatively assessed included threshold, latency, amplitude, certain indices of fiber refractoriness, and the nature of the response waveform. All properties of the unitary responses were catalogued for correlation with respective properties of other thalamic units as described in the literature (26,54). Neurons were identified as belonging to a particular nuclear group based upon stereotaxic coordinates and on physiological properties. Histological techniques to verify unit location, although very desirable, could not be included for economic reasons.

Once a pulp-driven unit had been identified and the characterization of its physiological properties completed, definitive experiments were initiated. Control or baseline data in the absence of Acupuncture stimulation was collected, in which both the responsiveness to pulp stimulation and the nature of spontaneous activity of the thalamic unit was recorded. Control (baseline) data was collected for prolonged periods of time (up to 1.5 hours) in some experiments, to permit documentation of the consistency and stability of the experimental preparation. Once the control data had been collected, Acupuncture stimulation was initiated (waveform described above).

Acupuncture stimulation was continued for approximately 0.5 hours to allow for prolonged induction periods noted in certain experimental procedures in the literature (7,60). Upon termination of Acupuncture stimulation, the responsiveness of the pulp-driven thalamic unit was monitored for an extended time interval (minimally, one-half hour) to document the recovery process.

Data Analysis and Interpretation. As mentioned, the field-potential data was used to delineate regions of pulp-driven thalamic activity for consideration in later single-unit experiments. The data from single pulp-driven units was analyzed to classify their properties relative to other thalamic units (26,54), to provide a direct comparison of the nature of nociceptive units from tooth pulp relative to pain units from other sources and to provide a basis for interpretation of the Acupuncture data. Data from the prolonged control (baseline) episodes was quantitatively evaluated to assess the stability of our animal preparation in order to verify the validity of the procedure. Data collected during the application of Acupuncture stimulation was used to quantitatively compare unit behavior to the baseline data, with regard to both spontaneous and stimulus-evoked activity. The data from the recovery period, collected following the termination of the Acupuncture stimulation, was analyzed in a similar fashion. The data was interpreted not only in the light of the relative specific responsiveness during the different experimental periods, but also in terms of the known requirements for temporal and spacial summation associated with integrative processes in pain pathways. Definitive experiments were completed on eight PO units and three ML units.

Results

Field-Potential Studies. The field-potential mapping studies were conducted to delineate anatomical limits of pulp-driven thalamic activity for future investigations. Thirty five field-potential experiments were completed. Pulp-driven activity seemed to be localized to three distict areas centered at A6.0, L6.0, and H+3.0 (PO), A8.0, L5.0, and H0.0 (VB), and A7.5, L3.0, and H+1.0 (MT). These cooridinates were referenced to a point lying in the stereotaxic plane midway between the earbars. These limits were somewhat crude due to the macroelectrode used in the field-potential studies, but the correspondence with the particular nuclear groups was confirmed using preliminary experiments in the single-unit studies described below.

Physiological Properties of Thalamic Units. Using single-unit recording techniques, we have recorded from 178 diencephalic units. All units have been found to lie within $\pm$ 1 mm of one of the three thalamic nuclear groups of interest, based upon stereotaxic coordinates. The purpose of these experiments was to quantitatively survey the physiological properties of units in the three thalamic nuclear groups and to catalogue these general properties. This data formed the basis for the quantitative assessment of the efficacy of Acupuncture stimulation.

The physiological properties of single pulp-driven unit responses derived

from a generalized survey of all three thalmic nuclear groups have consisted of a single spike or a short burst of spikes (Table I). The spikes have been small (40-300 μv), and usually consisted of a negative phase followed by a positive phase of lower amplitude. A few positive-negative and positive-negative-positive spikes were also found but no purely positive spikes were observed, indicating that the responses were recorded extracellularly from cell bodies rather than axons (61). Latencies were mostly in the range of 15-30 μs and generally varied within 10 percent of the mean value for a particular unit. Latencies, burst sizes, and threshold to bipolar pulpal stimulation for the units found in MT, PO, and VB are given in Table I. Many individual units responded to stimulation of the mandibular canine ipsilateral to the test tooth (spatial convergence) and to tapping of the tooth, mechanical stimulation of the hard palate, or to the stimulation of other orofacial areas (polymodal convergence). In general, train stimuli as opposed to single pulses were much more effective in eliciting responses from the pulp-driven thalamic units.

Definitive Acupuncture Data from PO. As described in the Methods section, use of the dot-raster recording technique permitted the presentation of a large volume of data in a condensed and easily visualized form. Details of the dot-raster concept and the mechanics of its operation are also discussed in the Methods section of this report. In the experiments reported below, a dot-raster display was generated every five minutes. Within each dot-raster display, one stimulus trial was initiated every second, and each dot-raster display represented 64 stimulus-response trials. The dot-raster displays were generated only at five minute intervals, as the data would otherwise have been overwhelmingly voluminous.

Upon completion of the baseline characterizations of the physiological properties of thalamic units, definitive studies were initiated using Acupuncture protocols. Information from the general literature associates units of PO and MT with the motivational-affective dimension of pain, whereas units of VB are predominantly associated with the sensory-discriminative dimension of pain (45,46). Based upon this conceptualization, it seemed most logical to conduct the definitive experimental series looking at units of the various nuclear groups in the order of PO, MT, and finally VB. At the time of contract termination, we had obtained a preliminary definitive survey of units in PO and had initiated similar studies of units in MT. The physiological properties of the PO and MT units involved in the definitive experiments are summarized in Table II. We chose PO for the initial definitive series, because PO units were easily located and held for prolonged periods of time. Also, this nuclear group has been associated with aspects of pain other than pure somatotopic location (33,45-47). PO seems more likely associated with temporal summation and polymodal responsiveness, characteristics which are important to the affective or "hurt" component of pain.

Figure 1 illustrates the nature of the raw and summarized data for one particular PO unit. The raw data is indicated as three dot-raster displays occupying the left-hand column. As described in the Methods section, the experimental format was composed of a control period (top), a period of

TABLE I

Physiological Properties of All Thalamic Units Examined in Detail

Nucleus	PO (N=30)	MT (N=18)	VPM (N=31)
Mean Latency (msec)	18.40	28.22	30.14
Mean Number of Spikes in a Burst	2.4	3.1	2.9
Mean Threshold (μA)	140*	250*	170*

* Mean Threshold values based on sample sizes of 24, 8, and 7, respectively. Earlier data for these values were disregarded due to electronic recording which invalidated the results.

TABLE II

Physiological Properties of the Thalamic Units Used in the Definitive Simulated Acupuncture Experiments

Location	Latency (ms)	Threshold (ua)	Spikes/Burst
PO	15	140	2
PO	12	100	3
PO	8	30	5
PO	16	18	3
PO	14	30	3
PO	12	20	2
PO	15	60	4
PO	14	325	3
CM	14	100	4
CM	16	175	4
CM	12	40	4
$\overline{PO}$ (N=8)	13.3	90	3.1
$\overline{CM}$ (N=3)	14.0	105	4.0

The indicated value for the number of spikes per response burst for each PO unit was determined by observing a large number of stimulus-response trials and selecting a number which was subjectively felt to be most typical.

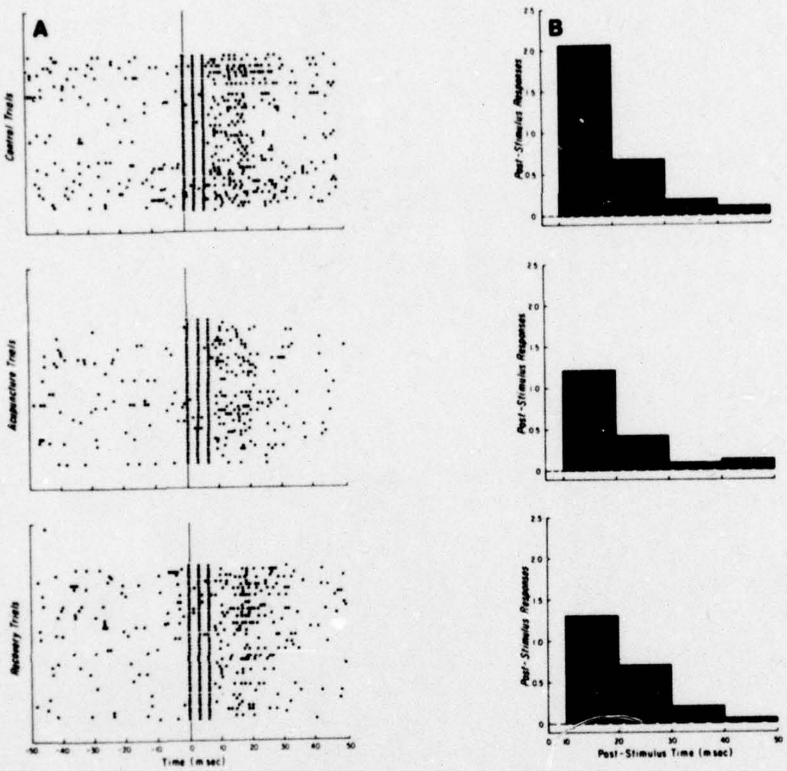

Figure 1 Dot-raster displays (A) and post-stimulus time-histograms (B) before (upper row), during (middle row) and following (lower row) simulated Acupuncture stimulation for one particular PO experiment. Control, simulated Acupuncture, and Recovery time periods were of 0.5 hr. duration. Test stimulation was initiated at zero time for each test trial; test trials occurred every second and are shown sequentially from top to bottom. An episode of 64 test trials was conducted every 5 minutes. The post-stimulus time-histograms do not include data for the first 10 ms time period because of the presence of test stimulation. The base of the histograms begin at zero because averaged spontaneous activity from the Control period was subtracted from the data.

Acupuncture stimulation (middle), and a recovery phase (bottom), each dot-raster display representing 64 stimulus-response episodes ordered from top to bottom. The upper display is an example selected as representative of the control or baseline period. Activity on the left half of the dot-raster display represents spontaneous activity prior to test stimulation. The tooth stimulus was applied at the center of the dot-raster display (a 10 ms train of three pulses; see Methods). In many cases, stimulus artifact intensities were within our window discriminator setting, and, as in the case depicted here, were registered as dots in a fashion similar to thalamic single-unit responses (three vertical columns of dots just to the right of the center of the display). During the baseline period, the data for this particular unit showed the typical burst-type response pattern of thalamic units, which then tapered off markedly after approximately 30-40 ms. During Acupuncture stimulation (center dot-raster display), the results indicated little change in spontaneous activity of the unit, but a significant decline (although not elimination) of the stimulus-elicited burst-type activity. Following termination of Acupuncture stimulation (lower display), the data indicated essentially full recovery of the burst-type activity, and, again, little change in the level of spontaneous activity relative to the baseline. Recovery was not always complete by the time of termination of the experiment, but the single-unit responses almost always showed a progressive tendency to return to the control level, and it appeared likely that recovery would have been complete if the experiment had been extended.

The right column of Figure 1 depicts time-histogram diagrams of the data summarizing the entire experiment. The height of each bar represents the number of times (on the average) the unit fired within the indicated post-stimulus time interval (10 ms time bins). The zero level on the graph does not represent the total lack of activity, but represents the fact that the average level of spontaneous activity for the control period had been subtracted from the data. The upper diagram represents the baseline period, and documents the initial stimulus-driven burst-like activity and the subsequent progressive tapering of single-unit response frequency to baseline. The middle diagram represents results during Acupuncture stimulation, and shows a significant decrease in both the frequency and duration of burst activity. Finally, the lower diagram represents the summarized results during the recovery phase, which in this particular experiment, reached an intermediate level of activity over the recording interval examined. It is important to note that initial trials immediately following Acupuncture stimulation were found to exhibit a significant attenuation of responsiveness, so the lack of full recovery in the data averaged over the entire recovery interval (and therefore including the initial trials) is not particularly surprising. The results of Figure 1 were generally typical of all experiments, although, in some cases, the level of recovery in the averaged data was more complete.

Figure 2 is a Time-Histogram plot summarizing all data from eight PO experiments in which the entire data collection protocol was completed. The format of the figure is identical to the summarized data of Figure 1, with the control period at the top, the Acupuncture episode in the center,

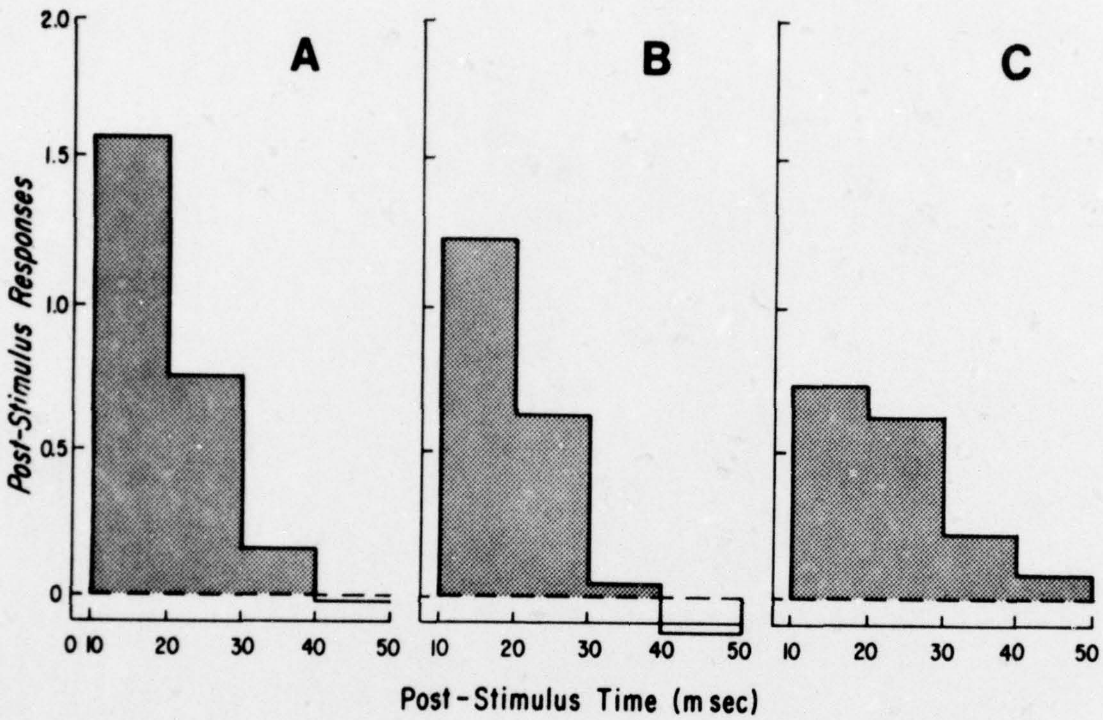

Figure 2 Post-stimulus time-histograms of data averaged over 8 PO experiments before (A), during (B), and following (C) simulated Acupuncture stimulation. Control, simulated Acupuncture, and Recovery time periods were of 0.5 hr. duration, except some Control sessions ranged to as long as 1.5 hrs. to verify preparation stability. Data was not included for the first 10 ms post-stimulus interval because of the presence of test stimulation. The base of the histograms begins at zero because averaged spontaneous activity from the Control period was subtracted from the data.

and the recovery period depicted in the lower portion of the figure. The summarized data of all experiments was similar to the example from a single experiment given in Figure 1. The data from the control period indicated a stimulus-induced high-frequency burst following stimulation, which then tapered to baseline responsiveness over a period of approximately 30-40 ms. Acupuncture stimulation again imposed a significant attenuation of burst-type activity, although not to the extent seen in the single experiment of Figure 1. Also, the single-unit activity was even more depressed during the recovery phase than during Acupuncture stimulation. This was not the result of overall deterioration of the experimental preparation, as documented in a later section. It was a manifestation of a continuation of depression in single-unit activity exhibited during the late stages of Acupuncture stimulation, and the fact that the effects of such stimulation seemed to greatly outlast the Acupuncture stimulation interval.

As mentioned previously, three definitive experiments were completed for units in MT. The MT tests involved the same Control-Acupuncture-Recovery experimental format as was used for the PO experiments. The left column of Figure 3 illustrates the nature of the raw dot-raster data from one of the three MT experiments completed, in a fashion analogous to the PO data format of Figure 1. Data from the control period (top display) showed the typical burst pattern of thalamic unit responsiveness, exhibiting a high frequency at first which then tapered to baseline levels in 40-50 ms. During the Acupuncture episode (center display), a slight increase in spontaneous activity was seen, but, in spite of this, there was a significant decrease in stimulus-driven burst-type activity. Following the termination of Acupuncture stimulation, in this example, the data indicated a continuing slight increase in the level of spontaneous activity, while the stimulus related responsiveness of the unit showed full recovery to control conditions. The other MT experiments gave qualitatively similar results, with a decline of stimulus-driven activity during the Acupuncture period and, following Acupuncture termination, a marked tendency for progressive recovery to control levels.

A summary of the entire results for the MT experiment described above is displayed in the right column of Figure 3 in Time-Histogram format similar to that of Figure 1. The baseline period showed the initial stimulus-driven high-frequency burst activity, which then declined in a typical fashion to baseline levels in approximately 40-50 ms. During Acupuncture stimulation, the data exhibited a significant attenuation in both burst frequency and duration. Finally, during the recovery phase, the single-unit responsiveness reverted nearly but not completely to the baseline level. As with the PO data, recovery was progressive, and later stages exhibited control-level responsiveness. The attenuation in PO responsiveness during Acupuncture or recovery could not be attributed to preparation deterioration, as documented below.

Baseline Stability. The question of preparation or baseline stability was of primary concern, because all quantitative interpretation was to be based on this assumption. Baseline stability referred to the reproducability of the threshold of the pulp-driven thalamic unit over extended periods of

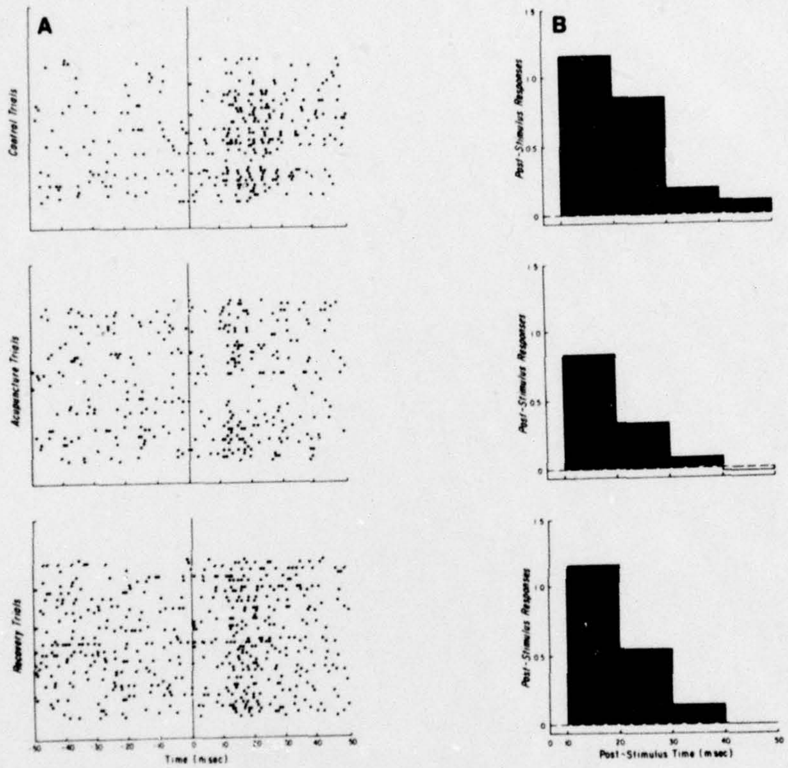

Figure 3 Dot-raster displays (A) and post-stimulus time-histograms (B) before (upper row), during (middle row) and following (lower row) simulated Acupuncture stimulation for one particular MT experiment. Control, simulated Acupuncture, and Recovery time periods were of 0.5 hr. duration. The experimental protocol and details of data presentation are identical to that of the PO data of Fig. 1.

time when unencumbered by Acupuncture stimulation. The problem of baseline stability was examined by conducting control sessions of extended durations. In general, the duration of control sessions was extended to as long as 1.5 hours with repeated testing of the threshold of the particular thalamic unit under investigation. The data indicated that the threshold of pulp-driven thalamic units was quite stable relative to the magnitude of changes induced during Acupuncture stimulation, as quantitatively documented in an earlier report (18), so that the experimental model had sound validity. Furthermore, these results were obtained at widely varying intervals following the initiation of the experiment, due to large variations in the time required to set up the electronic instrumentation and to locate and characterize a particular pulp-driven thalamic unit. Also, in preliminary experiments, it was qualitatively observed that individual units could be held for many hours with only minor alterations in response magnitude. Finally, a battery of general physiological parameters were routinely monitored during the experiments, and no significant changes in the physiological state of the preparation were observed. Therefore, changes seen during Acupuncture stimulation episodes or recovery periods could not be attributed to general preparation deterioration, to unit deterioration, or to progressive loss of the particular thalamic unit due to electrode movements, and the observed alterations in responsiveness could therefore truly be attributed to the particular form of Acupuncture stimulation employed.

Discussion

The data demonstrate that single pulp-driven units can be identified in three distinct nuclear groups of the thalamus, as determined by standardized extracellular single-unit recording techniques. The locations of individual units were identified on the basis of physiological characteristics and stereotaxic locations. The PO units studied exhibited a mean latency of 18.4 ms, a threshold of 140 μa, and an average burst duration of 2.42 spikes. MT units showed values which were 10-20 percent higher than PO units in each category, while VB units exhibited the longest latencies and burst durations, but intermediate thresholds. Upon imposition of definitive Acupuncture protocols, the majority of the eight PO units and the three MT units which were studied showed a significant and progressive attenuation of stimulus-driven responsiveness during Acupuncture, an effect which greatly outlasted the duration of the Acupuncture stimulation. Recovery was progressive, and in many cases, after 40-50 minutes, had returned substantially to control levels. These changes cannot be attributed to loss of the unit or deterioration of the preparation in general, as verified by the baseline data collected over extended intervals. Usually, the recovery periods were not of sufficient duration to permit the demonstration of complete recovery of the unit, but during the entire recovery period, the majority of units exhibited a marked progressive tendancy to return to control values. It seems reasonable to assume from the data, that had longer recovery intervals been included, many of the latter units would have tended to further return to baseline conditions. The spontaneous activity of the PO and the MT units was not significantly affected, in most cases, by the form of Acupuncture stimulation employed.

The observed pulp-driven thalamic units exhibited small amplitudes and relatively long latencies. These results probably indicate that the cells producing these responses were small. Pulpal primary afferents are known to be principally of the A-delta class (20,23,31,62), and this trend toward small cells seems to be carried on to the level of the thalamus. Thresholds for pulp-driven activity were quite high in the thalamus, and the use of train stimuli was nearly always obligatory. This contrasts with the lower thresholds using short single pulses which we have shown to be sufficient for excitation of pulp-driven units in the Gasserian ganglion (39,41) and brainstem Trigeminal Complex (63), and emphasizes the necessity of either temporal and/or spatial convergence for excitation at the level of the thalamus. Data from the work of others supports the same conclusion (28, 43).

The spatial and multimodal convergent properties of the pulp-driven thalamic units examined to date were not unexpected, as these properties have long been assocaited with these thalamic centers (25,26,54,46,64). However, units of VB have classically been considered the antithesis of these properties (25), having a precise somatotopic organization in which convergence from the upper and lower jaw would certainly not be expected. It appears, then, the pulp-driven units we have found in VB belong to the recently discovered subset of ventrobasal cells with "extra-lemniscal" properties (26). In common with extra-lemniscal cells, they show spatial and temporal convergence, low following rates to repetitive stimulation, and small size.

Our conceptual approach in the Acute Neurophysiological Experiments is adequately supported by the data as presented. The most comprehensive model of pain proposed to date describes three ascending pathways which directly dictate or modulate the painful experience (45,46), the pathways all having major relays possessing significant integrative responsibilities at the level of the thalamus. The anatomical proximity and integrative functions of these systems ascribed to the thalamus make this level an extremely logical point to define an index of nociceptive activity indicative of analgesic effects. Therefore, there is every reason to suppose that our initial hypothesis regarding experimental design was accurate in terms of general experimental knowledge and theoretical constructs and the present experimental data. Furthermore, the data directly demonstrate the feasibility of the model, and, to the extent that definitive data was collected, demonstrate that the form of Acupuncture stimulation which was employed definitely exerted significant effects. The described model therefore exhibits all properties sufficient to have permitted the ultimately sought identification of a central nervous system "index" of Acupuncture analgesia effects, and the latter index would have provided the means to quantitatively optimize Acupuncture administration strategies in the Acute experimental model.

CHRONIC NEUROPHYSIOLOGICAL EXPERIMENTS

Perhaps the most powerful experiments available for investigation of analgesia are Chronic (Behavioral) Neurophysiological Experiments, because they permit the evaluation of central nervous system functions free from complications introduced by anesthetic agents and surgical intervention in Acute experiments and they permit more flexibility than studies in humans. Furthermore, as a component of the present research program, the Chronic experiments provided the main framework as a link between data accumulated in the Acute versus the Human experiments. Chronic experiments were of further value in this particular case because they permitted evaluation of Acupuncture effects free of placebo reactions, suggestion, or hypnosis, and provided the flexibility of imposition of experimental conditions which would not have been permissible for ultimate human studies. They were also crucial to verify information derived from both Acute and Chronic experiments at the actual level of perception and to obtain data crucial for documentation of safety. The central feature of the Chronic Neurophysiological Experiments was the development of a model which employed perceptual rather than reflex manifestations of sensation as an index of Acupuncture effectiveness. To adequately study the efficacy and feasibility of Acupuncture, a model was required in which the animal could report pain thresholds (based on perceptual criteria) on a continuous basis over an extended period of time.

The Chronic Neurophysiological model had as its central features the chronic implantation of electrodes in a tooth pulp to permit application of noxious stimuli, the chronic implantation of electrodes at specific Acupuncture points to permit study of analgesic efficacy of such stimulation, the implantation of a head pedestal allowing electrical communication with external apparatus, and the use of a perceptual (as opposed to a reflex) index of pulpal perceptual sensibility.

Noxious stimulation was initiated by activation of tooth pulp afferents for the same reasons as detailed under the description of the Acute Neurophysiological Experiments. Tooth test stimuli were administered to the same tooth (maxillary canine) and in the same manner (0.1 ms rectangular pulses in the bipolar stimulus configuration and a train-stimulus mode) as that used for the Acute experiments. These particular features of the Chronic and Acute experimental models were duplicated to optimize interpretive overlap between the two experimental programs. Further details of the noxious stimulation system are given under the Methods section.

The site of Acupuncture stimulation was selected initially to be the Hoku point on the forelimb, again, for the same reasons as detailed for the Acute Neurophysiological Experiments. Furthermore, the Acupuncture stimuli were administered in the same fashion (electrical stimuli administered through a wire wrapped around the superficial branch of the radial nerve) and using similar electrical waveforms to those employed in the Acute experiments to provide maximum overlap with the latter experimental model.

All experiments were conducted using standardized psychophysiological stimulation and recording techniques, and employed the cat as the experimental animal for economy and for maximal correlation to the general pain control literature (20,24-35), previous work in our laboratory (36-42), and for maximal correlation with the Acute experimental model. The particular psychophysiological model chosen utilized the Threshold Titration paradigm, an experimental procedure permitting the quantitative monitoring of perceptual sensibility over extended periods of time. A detailed description of the Threshold Titration paradigm is presented below.

The investigation of all Acupuncture points relevant to dental analgesia was originally scheduled within the present experimental program. However, the premature termination of the project prevented the collection of extensive definitive data; a few preliminary experiments related to the Hoku Acupuncture point qualitatively indicated an increased tolerance to noxious pulp stimulation (results which were analagous to those of the Acute Neurophysiological Experiments). The major data to be presented documents the operational status of the complicated training procedures, the Threshold Titration paradigm, and the requisite experimental equipment and facility which comprise the Chronic Neurophysiological model as developed.

Methods

Animal Preparation and Surgery. Experimental animals were initially screened for suitability and subjected to preliminary training using a noxious footshock program (see below), and were then scheduled for surgery. Endotracheal intubation was accomplished using a short-acting barbituate. Surgical anesthesia was induced under Ethrane-Oxygen and maintained using Ethrane-N_2O-Oxygen. Subsequently, an optically-coupled ECG (to preserve ground isolation) was attached (Terrasyn, Model N-IIIB ECG Isolation Amplifier), and a non-invasive blood pressure monitoring system was applied to one of the forelimbs (Hoffman-La Roche Arteriosonde, Model 1010). In addition, a Bechman LB-2 Medical Gas Analyzer, sampling the respiratory gases, was employed to continuously monitor end-tidal CO_2 as an index of short-term acid-base balance. Respiration was assisted on demand, or controlled, using a Bird Mark IV-VIII Anesthesia Assistor. Intraesophogeal temperature was monitored (YSI Model 43TA) and maintained at 38 $\pm$ 0.5 C by means of a heating pad with thermostatically controlled circulating water (Gaymar Temp-Pump System). The animals were then brought through the recovery process, and allowed several days to recuperate from the surgical procedure prior to the initiation of experiments. Long-term health status was documented through periodical physicals.

Noxious stimulation was accomplished through electrodes placed in the experimental tooth (maxillary canine) (24,37). One electrode was placed in each of two cavities drilled through the enamel to a near pulp exposure. The base of each cavity was filled with silver amalgum, the electrode wire (stranded stainless steel) was packed in place, and the whole complex was sealed in position using non-conductive adhesive. One of the tooth stimu-

lation electrodes was located on the lingual aspect and one on the buccal aspect of the tooth, both being near to but absolutely distinct from the gingival margin. The wire leads (teflon coated) were implanted under a muco-gingival flap, and then directed upward under the facial skin along the surface of the bone between the medial canthus of the eye and the nose, to gain access to an electrical connector mounted in the head pedestal (24). Initial training sessions involved footshock as an alternative form of noxious stimulation, but no surgical procedures were required.

Acupuncture stimulation was accomplished using electrodes implanted by techniques developed in another laboratory. In this procedure, the electrode wire (bared for 0.5 cm) was carefully wrapped around the appropriate nerve (superficial branch of the radial nerve just distal to the Acupuncture point for Hoku) and sewn to nearby muscular tissue. The Acupuncture lead was directed subcutaneously from the site of nerve placement to the anterior surface of the head, for attachment to the electrical connector of the head pedestal. Stress imposed by joint or muscular movements was minimized using carefully defined courses for the Acupuncture leads.

The head pedestal to which the tooth and Acupuncture leads projected was built up by repeated applications of dental acrylic, using methods adapted from a report in the literature (24). The acrylic pedestal was held in place by molding it around stainless steel screws mounted in the skull of the animal. The lead wires were connected to a multipin electrical socket, which was then embedded in the acrylic pedestal. A flexible cable connected the pedestal plug to a multi-contact commutator suspended from a counter-balanced pulley system to provide minimal restrictions of vertical and rotational movements of the animal.

Stimulation Techniques. Three stimulus situations were involved in the Chronic experimental model, that of footshock, pulp stimulation, and Acupuncture stimulation, respectively. Special considerations related to each of these techniques are presently discussed.

Footshock of noxious intensities was used for initial training procedures (see below). Footshock consisted of the application of stimuli to aluminum bars in the lower portion of the cage at intensities determined to be definitely aversive independently for each animal prior to each experimental session (typically less than 0.2 ma). The stimulus waveform was a 60 ms burst of 4 equally-spaced rectangular pulses of 10 ms duration, with a burst repetition rate of 10 Hz.

The important consideration relative to tooth stimulation was that of requirements for temporal summation of activity in pain pathways, a phenomenon recently realized to be important for nociceptive interpretation in the central nervous system (45-47). A train stimulus format was therefore adopted, to induce an activity profile in pulpal afferents which was somewhat dispersed in time. Based upon our previous experimental determination of the Strength-Duration curve for pulpal afferents (37), we chose 0.1 ms

for the width of each rectangular stimulus pulse. The frequency of stimulation was selected such that the majority of pulpal afferents could faithfully follow each stimulus pulse, based upon our data from single pulp-driven units in the Gasserian ganglion (41,42). The latter data indicated that the majority of the units responded faithfully to frequencies up to 300 Hz. Such high frequencies were also chosen to accentuate perceptual as opposed to reflex responsiveness (65). Based upon these considerations, the waveform chosen for pulpal stimulation was a burst of four rectangular pulses of 0.1 ms duration at a frequency of 333 Hz.

Requirements for Acupuncture stimulation were entirely distinct from those related to tooth pulp stimulation. A variety of waveforms have been described in the literature for both human and animal studies (7,9,17,49). Waveform was obviously identified as a very important variable for investigation, and many waveforms were scheduled for eventual investigation. However, for initial tests of feasibility, a train of 0.1 ms rectangular pulses at a frequency of 50 Hz was employed, because similar waveforms have been shown to work in both chronic animal (14,16) and human (7,17,49) studies. The proper intensity for Acupuncture stimulation was again difficult to determine, but was obviously another variable of considerable importance. For our preliminary tests in the Chronic Neurophysiological Experiments, we chose to use an intensity somewhat below that which induced muscle fasciculations visible to the naked eye and, which, in the behaving animal, did not produce visible signs of perceptual discomfort.

Training Program. The desired Chronic Neurophysiological model had as its central feature the use of a perceptual index to monitor pulp threshold, the Threshold Titration paradigm of physiological psychology (66). The complicated nature of the latter procedure necessitated designing a training program in which the animals were conducted through several preliminary behavioral stages which successively approximated the desired end-result. As an initial step, the animals were subjected to a footshock paradigm in which noxious footshock could be terminated by pressing a lever. With experience, it became possible to identify animals which would be successful in later stages of training based upon this initial footshock session. Animals deemed acceptable were then subjected to additional footshock sessions until their performance reached a level in excess of 90 percent escape behavior (typically, 3-5 sessions). The animals were then taken to surgery, and the pulpal and Acupuncture electrodes, the lead wires projecting to the top of the head, and the head pedestal with its embedded electrical connector were implanted. Following recovery from surgery (2-3 days), the animals were subjected to at least one additional footshock session to verify retention of previous training. The animals were then introduced to the next stage of training, in which the site of noxious stimulation was shifted from the footpads to the tooth pulp, with all other parameters of the experimental paradigm unchanged. Pulp stimulation (waveform previously described) was presented at a rate of 2.5 Hz and at an intensity determined to be definitely aversive prior to the training session. A transfer of trianing from footshock to tooth pulp proved to be a relatively straightforward generalization process, and the animals became proficient

at greater than the 90 percent level of escape behavior in 2-4 sessions of pulp stimulation.

Once training to the tooth pulp stimulation paradigm was deemed sufficient, the animals were introduced to the final procedure in the experimental series, the Threshold Titration paradigm (66). A subthreshold intensity was used initially and repeated a given number of times (selectable), but following every N'th (usually 8) succeeding stimulus, the intensity was automatically incremented to a new plateau. Stimuli (waveform previously described) were presented at a rate of 1 Hz. In the absence of corrective behavior by the animal, this process continued with equal intensity increments after every N'th stimulus until noxious or potentially noxious intensities resulted in immediate bar-pressing responses, thereby resulting in a lowering of the stimulus intensity to zero. Our results demonstrated that the animals tended to allow the intensity to climb to a relatively stable level during each ascending episode, this behavior being observed for prolonged periods, results which were consistent with reports in the literature (66). This described procedure obviously provided a direct quantitative measure of perceptual responsiveness (threshold) to noxious stimulation over extended periods of time, an extremely powerful experimental model. Finally, once the animals were deemed proficient in the Threshold Titration paradigm, Acupuncture stimulation protocols were introduced to test the effect on perception of such procedures. As of the date of project termination, several such procedures could be repeated daily (in different animals), permitting the accumulation of a voluminous amount of data in a relatively short time period and thereby greatly reducing the number of animals and time commitment required to generate statistically significant conclusions.

The various procedures associated with the Chronic Neurophysiological Experiments were accomplished in a specially designed experimental cage, allowing for footshock and access to the pulp and Acupuncture stimulating circuits, while still permitting maximal animal mobility. The experimental cage was also designed to eliminate the animal's use of the walls to avoid footshock, to counteract the contingency of the animal continuously holding down the bar rendering the procedure ineffective, and other experimental problems. The entire experimental protocol, involving the presentation of stimuli and recording of responses were controlled and recorded entirely automatically for all training procedures and for the Threshold Titration paradigm. Automated behavioral training is termed "Automatic Shaping" (67), and, as described in detail below, is an experimental procedure which permits substantial reductions in personnel commitments. A microprocessor has also been recently introduced to control the Automatic Shaping procedure, replacing antiquated electronic control equipment employed previously.

At the time of program termination, all elements of the experimental facility, training procedures, and the Threshold Titration paradigm were operational.

Experimental Protocol. The Chronic Neurophysiological Experiments involved three successive stimulation techniques, footshock, pulp stimulation, and the Threshold Titration paradigm (the latter with or without accompanying Acupuncture stimulation).

For footshock sessions, the animal was placed in the experimental chamber and the Automatic Shaping equipment (described under Results) was engaged. This resulted in the presentation of a train of stimuli (waveform previously described) to foot bars in the lower section of the experimental chamber. The stimuli were continued for two minutes unless the animal pressed the response lever, at which time the footshock was immediately terminated. The footshock was derived from a constant-current generator (lab constructed) which supplied a given amount of current regardless of the number of foot bars that the animal happened to be straddling (68). A fixed rest interval was provided between repeated footshock episodes, and the total duration of the training session was 80 minutes. In general, the animals moved about aimlessly at first but very quickly developed an association between the response bar and the footshock. Following more trials, they became fairly proficient in escaping footshock stimulation once it had been initiated, even in the initial experimental session. A signal light and an audio tone were presented simultaneously with footshock to hasten training. Training sessions were continued until a level of 90 percent escape behavior had been attained. Animals were then identified as candidates for surgery, and the surgical procedure was scheduled. The results of our footshock system (successful acquisition of escape behavior in 3-5 sessions) compared very favorably to results reported in the literature, which indicates that 5-10 sessions are required for successful training in escape (69).

Following surgery, the animals were reacquainted to footshock and then introduced to the pulp stimulation paradigm. Pulp stimuli (waveform previously described) were presented under the same Automatic Shaping format as that used for the initial footshock selection procedures. The animals were trained in repeated sessions (80 minutes duration) during which continuous monitoring of escape responses or failure was recorded. A signal light and a tone were presented simultaneous to pulp stimulation in initial sessions to aid generalization, but in later sessions the tone and light were omitted to further shape behavior towards the ultimately sought Threshold Titration paradigm in which the signal light and tone were not employed. The criterion for success in the pulp stimulation paradigm was performance at the level of virtually 100 percent escape behavior, a situation usually realized after 2-4 experimental sessions.

Animals successfully trained in the pulp stimulation paradigm were then introduced to the Threshold Titration paradigm, individual sessions of which lasted for 80 minutes. As described previously, stimuli (identical to those used in the pulp stimulation training paradigm) were presented once per second with intensity increments introduced after every N'th (usually 8) stimulus. The approximate threshold of pulp stimulation was estimated at the beginning of each experiment, and the maximum intensity of the Threshold Titration unit was set so that the threshold of that animal

on that particular day occurred at roughly intensity step 3-5. At this point the microprocessor program was initiated, and the animal was maintained in the Threshold Titration paradigm for the 80 minute experimental interval. The data was continuously recorded as stimulus intensity versus time on a chart recorder. The animals could be used at a rate of one session per day, permitting the collection of a voluminous amount of data.

Data Analysis and Interpretation. The training data of the footshock and tooth pulp stimulation paradigms was in the form of total lever presses for various response possibilities of each stimulus-response episode, summed over the entire experiment (see Results for details). This data was analyzed in terms of percent responses in each category to initially evaluate the adequacy of the training procedure and eventually to verify the suitability and training progress of each experimental animal. The Threshold Titration data was recorded as the level of stimulus intensity which the animal tolerated for each stimulus episode and the length of time that that particular level was tolerated before the lever was pressed. At a given stimulus intensity, analysis of the time prior to a lever press did not prove to correlate in any way with behavior or stimulus intensity tolerance, and such data was therefore omitted from the quantitative results. Data relative to the stimulus intensity tolerated over the many stimulus trials in a given experiment was highly significant. The latter data was analyzed in terms of the actual intensity level and the degree to which variability was maintained within a given experiment, and comparisons of average intensity and variability profiles between different experiments in the same and different animals. As of termination of the project, data related to the administration of Acupuncture stimulation had only been qualitatively evaluated, and data eventually scheduled relative to physiological and psychological safety had not been collected.

Results

Development of the Facility. The total experimental facility for the behavioral experiments involved the experimental chamber, the electronics for automatic training, the footshock, tooth pulp, and Threshold Titration stimulators, and other accessory stimulation and recording equipment.

The experimental chamber underwent considerable design modification as accumulated data and experience dictated the necessity for alterations. Such modifications are detailed in earlier reports (18,53), and included alterations to eliminate "safe spots" or opportunities for the animals to minimize shock effectiveness, to allow for the efficient handling of waste products, apparatus to allow for virtually unlimited rotational and vertical movements of the animal, modifications of the chamber and sequencing electronics appropriate to handle the contingency of the animal's laying continuously on the bar, and several other more subtle contingencies. The final version of the chamber was described in considerable detail in the most recent annual report (18). Automatic Shaping procedures were used from the very beginning of the Chronic Neurophysiological Experiments. We initially used an automatic timing and sequencing equipment array on loan from NASA. Specific automatic training protocols were continually

upgraded as a result of extensive preliminary experiments to develop the final training paradigms for footshock and tooth pulp stimulation. Later, the NASA equipment became unavailable. Although it would have been possible to replace the equipment with a commercially available automatic timing and sequencing system for psychophysiological experimentation, we chose to replace the system with a microcomputer. The latter system permitted considerable savings in cost while simultaneously providing for greatly increased experimental flexibility and reliability. At the time of project termination, the microcomputer system and all requisite interfacing to the other experimental equipment was completely operational.

Three stimulation systems were employed in the Chronic experiments. For the Footshock training paradigm, a lab-constructed generator derived from information in the literature was used (68). The latter devise was completely self-contained and eventually was interfaced for microcomputer control. For the Tooth Pulp training paradigm, a Grass S-8 in the train or burst stimulation mode was employed, and appropriate interface circuitry was also introduced to place this instrument under control of the microcomputer. For the Threshold Titration paradigm, a suitable generator was designed and fabricated based upon our specifications in conjunction with electronic engineering consultation. The latter device was modified periodically to optimize performance based upon preliminary experiments and finally to provide for microcomputer control. Details of the Threshold Titration Generator have been presented in previous reports (18,53).

Experimental Results. Five categories of experimental results are presented, Automatic Shaping, a description of the various facets of the selection and training procedures in their fully developed form, data documenting behavioral modification from footshock to pulp stimulation as the aversive stimulus, data characterizing behavioral modification from pulp stimulation to the Threshold Titration paradigm, and the Threshold Titration and Acupuncture results.

In the Chronic Neurophysiological Experiments involving Automatic Shaping, two types of behavior were involved, escape behavior (the animal terminated shock once it was initiated) and avoidance behavior (the animal responded such that the stimulation was avoided altogether). In an individual experimental session which contained a series of stimulus presentation episodes, several time intervals were important: a), the inter-trial interval (ITI), the interval between the termination of the aversive stimulus and the beginning of the next stimulus trial; b), the unconditioned stimulus (US), the application of the stimulus of interest, in this case, the aversive stimulus; and c), the conditioned stimulus (CS), in the present case, a signal light and/or audio tone which warned the animal of an impending stimulus. The CS was used only when avoidance behavior was permitted. Avoidance behavior was permitted early in the overall program to evaluate the degree of learning during the development of training techniques, but was not used after routine training procedures had been established.

Operation and control of the conditioning periods were accomplished initially by the use of front-patched general control equipment for behavioral research (on loan from NASA) and, most recently, by the use of a microprocessor. Either component system was programed for an escape-avoidance schedule with limited hold (in the case of no response, the stimulus was turned off after a preset US interval). Either system was also programed to allow for various more subtle behavioral contingencies such as the animal laying continuously on the bar or finding safe spots, etc. Programed alternatives were also set up to permit selection of the Footshock training paradigm, Pulp-Stimulation training paradigm, or the Threshold Titration paradigm.

To summarize the training sequence, in the case of avoidance training, a trial was initiated with CS onset (the signal light and/or audio tone were presented in the absence of the noxious stimulus). If a response occurred within five seconds (avoidance behavior), the CS was turned off and the ITI interval was initiated (45 seconds duration). If no response occurred within five seconds, the US was initiated (while the CS remained on). Both the US and CS were then terminated by a response (escape behavior), or alternatively, by time-out of the US interval (one minute). In the case of escape training in the absence of the avoidance response possibility (a procedure used exclusively in the later periods of the program), the CS was bypassed, but otherwise the format was similar to the avoidance paradigm. If the animal was holding the bar down continuously at the initiation of CS (or US in the absence of avoidance behavior), and was still holding the bar down when the US interval timed out, the response bar was energized for a few seconds to back the animal off the bar (bar-hot interval). A subsequent bar press by the animal or time out of the bar-hot interval then led directly to another ITI interval. Experiments were automatically terminated at 80 minutes.

Data characterizing the Footshock training pardigm is presented in Table III. The data summarizes the percentage of escape responses for the first three sessions for many experimental animals; further sessions were not summarized because animals typically were deemed acceptable for further procedures after three Footshock sessions. The data dramatically demonstrate that the animals achieved remarkable levels of escape behavior even in the first experimental session. Animals moderately proficient in the first experimental session exhibited further increases in proficiency with additional experimental sessions, while animals demonstrating high levels of initial proficiency were seen to generally maintain their initial performance levels. With only two exceptions (animals number 66 and 67), all experimental animals were proficient at greater than the 90 percent level of escape behavior after three sessions. Experimental sessions were usually conducted on sequential or alternate days. Thus the data demonstrate the remarkably rapid training available using our particular adaptations of Automatic Shaping training procedures. The efficiency of training substantially exceeds anticipated performance levels described in the initial program proposal.

Table IV represents the raw data from Tooth Pulp training sessions for

TABLE III

Raw Data of Percentage of Trials in Which the Experimental Animal Exhibited Escape Behavior in the Footshock Training Paradigm

Animal Nr.	Session 1	Session 2	Session 3
46	92	100	100
48	100	96	90
49	88	100	99
50	100	100	95
53	79	97	92
54	87	89	92
55	69	59	100
65	82	88	100
66	100	86	75
67	85	96	82

TABLE IV

Raw Data of Percentage of Trials in Which the Experimental Animal Exhibited Escape Behavior in the Tooth Pulp Training Paradigm

Animal Nr.	Session 1	Session 2	Session 3
49	66	66	89
50	78	97	100
65	84	85	70

animals in which three experimental sessions were completed. These experiments were conducted following the surgical procedures and reacquaintance with the Footshock paradigm. It should be noted that the Pulp stimulation results can be correlated directly with Footshock data from the same animals (Table III). Fewer animals are represented in the raw data related to Tooth Pulp stimulation, as compared to Footshock stimulation, because of attrition related to microprocessor development, surgical complications, lack of definitive experimental data, and other reasons. However, as of the time of project termination, all of the latter contingencies had been reduced substantially. The raw data of the table indicates that the animals rapidly developed a high level of proficiency in pulp-induced escape behavior commensurate with the Footshock results of Table III. Additional animals in which fewer experimental sessions were completed also demonstrated similar behavioral profiles, and animal number 65 of the table, whose performance appeared to drop in session three, acquired a significantly higher level of proficiency in later experimental sessions. The raw data of the table and our experience to date demonstrate two facts. First, the transfer or generalization of behavior from the Footshock to the Tooth Pulp stimulation paradigm is efficient and rapid. Secondly, the animals respond with a high degree of efficiency of escape behavior following sufficient training in the Tooth Pulp stimulation paradigm. The temporal profile of acquisition of proficient performance in the Tooth Pulp training paradigm was unquestionably superior to expected performance levels as outlined in the initial project proposal.

An example of the raw intensity data of an individual Threshold Titration experiment is presented in Figure 4. Each bar depicts the results of an individual intensity stepping series, the bar height representing the intensity level at which the animal finally terminated stimulation with a lever press. Subsequent to each lever press, the intensity was returned completely to zero and another intensity stepping series was initiated. This particular experiment was selected because the results demonstrate several pertinent facts noted to differing degrees in various experiments. The average level at which the animal terminated stimulation remained approximately the same over extended periods of time. It was noted in some cases, however, and exemplified in the present figure, that the variability tended to change with time and sometimes became more pronounced near the end of the experiment, possibly reflecting periodic or eventual boredom or fatigue on behalf of the animal. This phenomenon is not believed to be the result of any Electroanalgesia effect resulting from test stimulation of the tooth pulp, based upon the results of our Acute Neurophysiological experiments (38-41). Finally, it is interesting to note that after many of the episodes in which a high intensity of stimulation was tolerated, the animal permitted only a very small intensity on the subsequent stimulus episode, a finding exemplified for some of the high intensity episodes in Figure 4. The data presented in Figure 4 is representative of all data collected to date, and unequivocally demonstrates the power of the present adaptation of the Threshold Titration technique for long-term threshold monitoring.

The data presented above strongly supports the feasibility of the Chronic

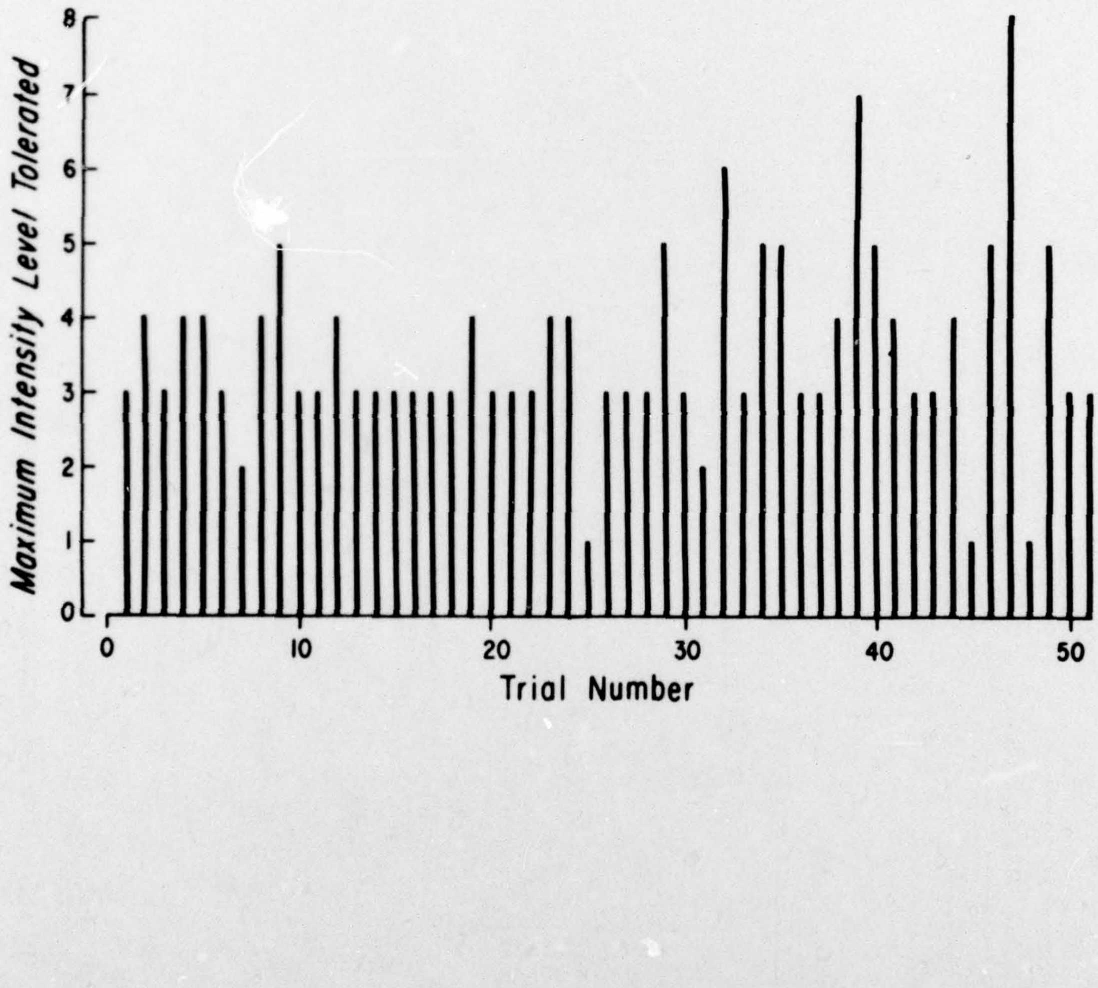

Figure 4 Raw intensity Threshold Titration data for one particular Chronic Neurophysiological Experiment.

pulp-stimulation model, substantiating data and conclusions from previous reports (18,53). Furthermore, the present data provides a powerful demonstration of the feasibility of the Threshold Titration paradigm. This was the major goal sought in the Chronic Neurophysiological experiments prior to the actual use of the model to evaluate Acupuncture. The latter step in the experimental series was only attempted in a qualitative fashion involving a few preliminary experiments prior to termination of the program. Nevertheless, the preliminary data definitely indicated that the application of Acupuncture stimulation of the form describe resulted in an increase in the maximal level of pulp stimulus intensity which the animal would tolerate. We thus conclude that the complete experimental model including both test and Acupuncture stimulation in a model permitting the monitoring of perceptual threshold over extended periods of time was developed to a completely operational status.

Discussion

All key goals regarding the experimental facility and the animal surgical and training procedures were accomplished. Complete electronic systems for Footshock, Tooth Pulp, and Threshold Titration stimulation systems were designed and constructed, and following modifications and the addition of a suitable recording system, the entire facility was brought to a status of complete readiness for definitive experimental procedures. Also, Automatic Shaping equipment was acquired, procedures were developed, and eventually the system was modified for control by a microcomputer. In addition, a complex experimental chamber was evolved through many stages of development, a devise which allowed for multiple contingencies inherent in the various paradigms employed. Finally, all surgical procedures required for the chronic implantation of stimulation electrodes and their communication with the external world were developed to operational status. Specific procedures allowing preselection of appropriate animals were also developed and incorporated into the experimental system. At the time the contract was terminated, the entire program of animal selection, surgical preparation, training, and execution of the Threshold Titration paradigm was operational and efficient.

The conditioning and training of cats to avoid an electrical shock by performing some response such as pressing a lever is a valuable technique for the study of motivation and behavior (70). At the same time, such avoidance and escape training has provided a useful model for measuring the sensitivity of animals to electric shocks administered through the foot pads (or other sources) and for observing the alteration of aversive thresholds through the application of various anesthetic or analgesic strategies (69). Typically, a procedure required for training the animals to perform the specific response (operant) is referred to as shaping, and involves the prolonged and systematic reinforcement of behaviors (by an experimenter directly observing the animal) which at first may be dissimilar to the desired operant, but may be required to orient the subject animal toward the lever or toward a particular area in the experimental environment. Successive minor behavioral alterations are then introduced by the experimenter which more and more approximate the desired response. Finally,

the actual operant itself is presented. This procedure is tedious and time-consuming, and often not compatible with the total experimental design (71). As many as 15 hours for each animal can be required for successive approximation training or "shaping" to escape footshock or other aversive stimuli by pressing a lever.

An automatic and much abbreviated method for escape and avoidance conditioning, which circumvents the problems inherent in the basic shaping procedure, is also available (67). Termed Automatic Shaping, this procedure eliminates the rigorous successive approximation format and significantly reduces the time necessary for conditioning. The experimental environment is rigorously structured such that the probability of eliciting the desired response from the animal is maximized. For example, if the interior dimensions of the response chamber and the location and site of the response lever are correctly determined, then on the first conditioning trial, the animal will display exaggerated motor activity in response to e.g. footshock, and this activity will lead to contact with the lever and termination of the shock. Following a brief number of trials, the desired operant is performed with regularity. Furthermore, and very importantly, this system can be automated. Particular aspects of our prior data, in some cases described in detail in previous reports (18,53), emphasize specific advantages of the Automatic Shaping program as adapted for our purposes. Firstly, the data has indicated that one session of Automatic Shaping to escape or avoid footshock is adequate to establish the association between a bar press and shock termination. Secondly, the performance level during the first experimental session can reliably be used to select suitable experimental animals prior to surgical procedures, thus reducing the risk of committing the time, personnel, and expense of preparation of an animal which, at best, would be difficult and costly to train at a later date. Furthermore, subjective observations provide definite evidence that there is a substantial decrease in jumping and struggling of the experimental animals during the initial test trials as the first session progresses. This is very desirable, because once the head pedestal and associated chronic implant lead wires are installed in the cat, exaggerated activity would greatly increase the probability of preparation damage. Also, a more sustained orientation toward the response lever is seen as the first session continues, and thus the animal is pre-conditioned to a more suitable position in the experimental chamber in preparation for later experimental sessions in the Footshock, Tooth-Pulp, and Threshold Titration stimulus paradigms.

The Threshold Titration paradigm is the ultimately desired experimental operant in our program and has been described in detail. The Automatic Shaping procedure described above has been adapted from basic considerations in the literature to peculiar aspects of our particular experimental situation which we have characterized experimentally. Our data indicates very efficient training, using the Automatic Shaping protocols, through all stages of the selection and training sequence to the final operant of the Threshold Titration procedure. Also, the Threshold Titration procedure has been demonstrated to work extremely well and to provide a powerful model suitable for studying various pain control strategies including the present

topic of interest, Acupuncture. The most striking aspects of the Threshold Titration paradigm are the sole dependence on a perceptual index of sensory sensibility and the ability to quantitatively measure perceptual thresholds over prolonged periods of time. The results of the program as of the date of termination showed that all aspects of the training and experimental procedure were completely operational. Furthermore, it was demonstrated that the microcomputer control and automation of data collection permitted the virtual full-time use of this model system on a daily basis with minimal personnel commitments. It is obvious that the original goals of the experimental program were achieved with admirable success, and, in turn, it is quite unfortunate that priorities dictated termination of the program on the brink of important definitive data.

SUMMARY AND CONCLUSIONS

1. Two definitive experimental programs were designed and developed to characterize the feasibility of Acupuncture Analgesia under conditions amenable to portable use in the field. The two experimental programs were designated the Acute Neurophysiological Experiments and the Chronic (Behavioral) Neurophysiological Experiments, respectively.

2. The Acute Neurophysiological Experiments were based upon recordings from single neurons in three thalamic nuclear integrative centers which have been implicated as having significant contributions to pain mechanisms, and the effects that concurrent Acupuncture stimulation protocols exerted on behavior of these thalamic units. The results included:

    a) development of an appropriate experimental facility and the delineation of specific areas of pulp-driven thalamic units;

    b) detailed characterization of the properties of single pulp-driven units in three distinct nuclear groups of the thalamus generally related to activity in pain pathways;

    c) definitive experiments to quantitatively characterize the effects of Acupuncture stimulation on pulp-driven unit activity in the two thalamic areas deemed most appropriate for the evaluation of Acupuncture Analgesia. The data indicated that Acupuncture stimulation of the Hoku point significantly attenuated such pulp-driven unit activity.

3. The Chronic Experiments were based on the Threshold Titration paradigm, in which experimental animals having chronic electrodes implanted in the tooth pulp (source of noxious activity) and near Acupuncture points were trained to regulate the level of pulp stimulation to intensities very near threshold for extended time periods. This permitted long-term monitoring of the efficacy of pain perception (not reflex responses!) during various control and Acupuncture administration protocols. The results included:

    a) development and documentation of appropriate facilities and surgical preparations for all facets of the proposed programs;

    b) development of three successive experimental paradigms culminating in the Threshold Titration program permitting long-term monitoring of perceptual threshold, using Automatic Shaping techniques under microcomputer control;

    c) documentation of the operational status of each experimental procedure and of successive transfer from one procedure to

another, and finally, using animals trained in the desired Threshold Titration operant, the demonstration that Acupuncture stimulation qualitatively increased the threshold to aversive stimuli as evaluated in the Threshold Titration paradigm.

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